National Park Service
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Natural Resource Stewardship and Science

Amphibian Monitoring Protocol (Version 2.0)

Great Lakes Inventory and Monitoring Network

Natural Resource Report NPS/GLKN/NRR—2018/1761



ON THE COVER
Gray Treefrog (*Hyla versicolor*) at Pictured Rocks National Lakeshore.
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Natural Resource Report NPS/GLKN/NRR—2018/1761

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Abstract

The National Park Service's (NPS) Great Lakes Inventory and Monitoring Network (GLKN) has developed a prioritized list of indicators, termed "Vital Signs", for monitoring long-term ecosystem health in NPS units in the Great Lakes ecoregion. This amphibian monitoring protocol is one of several protocols that GLKN is currently implementing is seven NPS parks: Apostle Islands National Lakeshore, Grand Portage National Monument, Indiana Dunes National Lakes Shore, Isle Royale National Park, Mississippi National River and Recreation Area, Pictured Rocks National Lakeshore, and Sleeping Bear Dunes National Lakeshore. Guiding principles for selecting this protocol's survey methods were: 1) maximizing detection probabilities, 2) minimizing cost, 3) practicality (in terms of non-experts being able to conduct surveys or collect data with minimal training and practical logistics), and 4) potential for partnering (in terms of using standard methods in widespread use to allow for data sharing)—all while meeting rigorous scientific standards for statistical power.

The program collects digital acoustic data utilizing automated recording systems deployed at wetlands within each park, supplemented with temperature loggers. This equipment is typically deployed in early spring, checked on once in late spring, and retrieved in August. Digital data are then archived and sent for analysis. The analysis consists of a series of steps designed to detect every species present at each site in each year with minimal false absences and no false positives. The latter condition is important for accurate occupancy trend analyses.

In brief, the analysis steps are:

1. Examine and organize annual data with quality control reporting.
2. Run acoustic samples (ca. 150/site/year) through software developed for automated species recognition.
3. Proof the auto-classification results to eliminate false positives for each sampling site.
4. Independently select 30 samples from the same dataset for manual surveys.
5. Manually survey these 30 samples, with an expert observer scoring each species present in each sample by visually and acoustically examining each acoustic signature encountered.
6. Have a second observer verify manual results.
7. Report on results.
8. Periodically (ca. every 5 years) run results through occupancy models to analyze trends.

This system provides an economically feasible methodology for the GLKN to track frog and toad occupancy trends within the region, and can be built upon incrementally to provide greater coverage as future needs dictate. It also allows for pooling of data with other regions or partners to expand analysis of natural resources monitoring, and the methodology can be readily developed further to monitor other wildlife that communicate acoustically (i.e., birds). Currently the program is monitoring 66 sites in seven parks, and analyzes approximately 2 TB of data annually.

Acknowledgments

This protocol benefitted substantially from a preliminary version completed by W. Sadinski (USGS Upper Midwest Environmental Sciences Center) and K. D. Bowen (National Park Service, Great Lakes Inventory and Monitoring Network), portions of which text were retained. We are grateful to J. Elias, B. Halstead, S. Hecnar, J. Mitchell, and B. Route for many fruitful discussions on monitoring feasibility and statistics. This work would not have been possible without extensive field work, and we thank all the park and other agency personnel and landowners who assisted with logistics and field surveys, sometimes even providing free housing and an occasional tipple. We also benefitted greatly from regional field work supported by the Lake Superior Binational Forum, the Ozaukee Washington Land Trust, the Great Lakes Indian Fish and Wildlife Commission, Trout Unlimited, and the Ottawa National Forest, enabling wider testing of automated recording systems. We are forever grateful for the many fine field biologists we have had the pleasure to work with, in particular R. and J. Rutherford. Development of this protocol was funded by National Park Service award P2105090018 to the senior author.

Revision History Log

Users of this protocol will promptly notify the program manager and/or the Great Lakes Network (GLKN) data manager about recommended and required changes. The program manager must review and incorporate all changes, complete the revision history log, and change the date and version number on the title page and in the header of the document file. For complete instructions, please refer to Revising the Protocol, SOP #9.

Previous Version #	Revision Date	Author (with title and affiliation)	Location in Document and Concise Description of Revision	Reason for Change	New Version #
1.0	23 Aug 2012	Gary Casper, GLES	p. i - Changed version # and authorship	Authorships finalized.	1.1
1.0	23 Aug 2012	Gary Casper, GLES	p. iv – added Acknowledgments section	Add acknowledgements	1.1
1.0	23 Aug 2012	Gary Casper, GLES	p. v – added revision history log line to TOC	Track revisions	1.1
1.0	23 Aug 2012	Gary Casper, GLES	p. vii – added Revision History Log section	Track revisions	1.1
1.0	23 Aug 2012	Gary Casper, GLES	2.2 – added subsection 2.2.1 Song Meter and Sensor Placement and Mounting	Add detail on equipment placement	1.1
1.0	23 Aug 2012	Gary Casper, GLES	2.3	Added clarification on calibration of SET programming to local time	1.1
1.0	10 Feb 2013	Bill Route, GLKN	Throughout	Made changes to ensure the protocol is consistent with USGS amphibian protocol	1.1
1.0	21 Feb 2013	Gary Casper, GLES	Throughout	Accepted Route changes, recalculated values in 4.4, 6.2 and 6.3, deleted scientific names from Table 2, updated common names throughout	1.1
1.1	2016	Gary Casper, GLES; Ulf Gafvert, GLKN	Throughout	Major revisions throughout after making sampling site selections, and finalizing data analysis protocols, including authorship.	2.0
Add rows as needed for each change or set of changes tied to an updated version number					

1.0 Background and Objectives

1.1 History

The National Park Service's (NPS) Great Lakes Inventory and Monitoring Network (GLKN) has developed a prioritized list of indicators, termed "Vital Signs", for monitoring long-term ecosystem health in nine NPS units in the Great Lakes ecoregion (Route 2004). This amphibian monitoring protocol is one of several protocols that GLKN intends to implement under the umbrella of an overall monitoring plan (Route and Elias 2007). Amphibians were initially considered for monitoring under a combined Amphibians and Reptiles Vital Sign. Because of differences in methods for monitoring both taxonomic groups and constrained funding and logistics, we decided to focus on amphibians initially with the intention of adding reptiles as the GLKN monitoring program matures.

Implementation of a viable, long-term monitoring program can be improved by partnering with other agencies, NGOs and universities around the Great Lakes region. As much as possible, implementation of this amphibian monitoring protocol will be a collaborative effort between the GLKN, the Upper Mississippi Region (UMR) of the U.S. Geological Survey's (USGS) Amphibian Research and Monitoring Initiative (ARMI) Terrestrial Wetland Global Change Research Network (TWGCRN; Muths et al. 2006), and other regional programs focusing on amphibian monitoring in the western Great Lakes (e.g., Lake Michigan and Lake Superior Lakewide Management Plans, Great Lakes Coastal Wetland Monitoring). In addition, state departments of natural resources in Indiana, Michigan, Minnesota, and Wisconsin use variations of the North American Amphibian Monitoring Program protocol (NAAMP; Weir and Mossman 2005). Similarly, the Marsh Monitoring Program is a joint Canada/USA effort to monitor amphibian populations around the Great Lakes (Crewe et al. 2006). The U.S. Forest Service and the U.S. Fish and Wildlife Service use variations of the NAAMP protocol to monitor amphibian populations on national forests and wildlife refuges in the region. As of this writing the NAAMP program has been discontinued but data collection utilizing the NAAMP protocol mostly continues by independent participants.

The sampling designs and methods described in this protocol are based upon our experiences surveying amphibians in the western Great Lakes region, and many fruitful discussions on practicality and statistical inference with network staff and other colleagues. We have conducted surveys in the Apostle Islands National Lakeshore (APIS), Isle Royale National Park (ISRO), Pictured Rocks National Lakeshore (PIRO), Sleeping Bear Dunes National Lakeshore (SLBE), Mississippi National River and Recreation Area (MISS), St. Croix National Scenic Riverway (SACN), Voyageurs National Park (VOYA), the Chequamegon-Nicolet National Forest (Moquah Barrens, Bayfield County, Wisconsin), Lake Superior Provincial Park (Ontario), the Thunder Bay region (Ontario), the Ottawa National Forest (western Upper Michigan) and throughout Wisconsin (Casper 2001a, 2001b, 2002, 2004, 2005, 2008, 2009, 2010a, 2010b; Bowen and Beever 2010; Casper and Anton 2008; Casper and Graff 2009; Hecnar and Casper 2009; Sadinski and Roth 2009). Conducting those surveys enabled us to test the survey methods we recommend in this protocol in a variety of habitats and conditions. Guiding principles for survey method selection for this protocol were, a) maximizing detection probabilities, b) minimizing cost, c) practicality (in terms of non-experts being able to conduct surveys or collect data with minimal training, and practical logistics),

and d) potential for partnering (in terms of using standard methods in widespread use to allow for data sharing) – all while meeting rigorous scientific standards for statistical power.

It is a near impossible task to balance these constraints for many secretive species, and the key to success is discovering species/survey method combinations that achieve high detection probabilities, and sampling many sites (Steen 2010). These useful combinations fell into two categories: calling frog surveys (most anurans), and visual searches for eggs (Wood Frog, ambystomatid salamanders, Four-toed Salamanders) or adult salamanders (Eastern Red-backed Salamander; Hecnar and Casper 2009). Some trapping methods were also highly successful (e.g., aquatic funnel traps for adult salamanders) but proved too costly to implement on a broad scale due to the time required for repeated site visits (Hecnar and Casper 2009). After due consideration of our guiding principles and discussions with network staff on feasibility, this protocol focuses solely on acoustic frog surveys using Automated Recording Systems (ARS), to both reduce the human workload, and to maximize sample sizes for achieving the best statistical significance possible. Acoustic surveys without ARS are in widespread use, and the data collected via ARS are expected to be comparable, allowing for widespread partnerships and data sharing. We also offer a specific standard operating procedure for timed visual searches as a supplemental option to monitor additional (mostly non-vocal) species.

We believe that additional survey methods with reasonable detection rates may yet be developed (e.g., eDNA sampling), which could improve or supplement this monitoring program. The methods presented here, if fully implemented with a sufficient number of sites monitored, will produce data that are statistically robust and comparable to results from other programs, including all programs that conduct acoustic surveys regionally and nationally and especially those implemented by ARMI and other NPS networks (Paton et al. 2003, Bailey et al. 2007a). These methods also supply high confidence for individual site monitoring, even when the number of sites sampled is insufficient for regional occupancy analyses.

1.2 Rationale for Selecting Amphibians

Amphibians were ranked among the top vital signs across the parks of the GLKN (Route 2004). Reasons to monitor amphibians include:

1. **Sensitivity**: Amphibians generally are sensitive to changes in environmental factors, including temperature, precipitation, humidity, hydrology, land cover, nutrients, toxicants and exotic species, among others (Sadinski and Dunson 1992, Blaustein et al. 1997, Rowe et al. 1998, Welsh and Ollivier 1998, Adams 2000, Pollet and Bendell-Young 2000, Gibbs and Breisch 2001, Marco et al. 2001, Sparling et al. 2001, Veldhoen and Helbing 2001, Beebee et al. 2002, Rowe et al. 2003, Johnson and Chase 2004). Recently, amphibians as a group have been ranked as only moderately sensitive to chemical contaminants compared to other taxonomic groups (particularly insects), but this finding does not obviate real declines and suggests that amphibians may be more sensitive to other factors (Kerby et al. 2009).
2. **Role as ecological integrators**: Most species of amphibians require both terrestrial and aquatic habitats, and their fitness is a function of environmental conditions across habitats. Pond breeding amphibians in particular illustrate the essential role of adequate terrestrial habitat to the maintenance of wetland biodiversity and ecosystem function, where amphibian

population persistence requires the maintenance of sufficient extent and quality of adjacent terrestrial habitat, and connectivity among local populations (Harper et al. 2008). Moreover, species with different life-history strategies respond differently to the loss and degradation of terrestrial habitat. For example, the Wood Frog, with a short life span and high fecundity, is more sensitive to habitat loss and isolation, whereas the longer-lived Spotted Salamander with lower fecundity is most sensitive to habitat degradation that lowers adult survival rates (Harper et al. 2008). Amphibians are also an important part of both aquatic and terrestrial food webs (Stebbins and Cohen 1995, Fauth 1999, Semlitsch 2000) and nutrient cycling (Milanovich and Peterman 2016).

3. **Ease of study**: Various life stages of amphibians are typically available and accessible for study. These traits allow for rigorous sampling designs for monitoring (Heyer et al. 1994, Fellers and Freel 1995, Corn et al. 2000). Amphibians also are invaluable for conducting manipulative experiments when further studies are necessary to establish the causes and effects of perceived declines (Sadinski and Dunson 1992, Boone et al. 2007).
4. **Conservation concern**: Declines of populations of amphibians are among the most prominent global issues in conservation biology (i.e., Young et al. 2001, Blaustein and Kiesecker 2002, Stuart et al. 2004, Lannoo 2005, Hoffman et al. 2010, Whitfield et al. 2016).
5. **Regional relevance**: All four states in which the GLKN is located, as well as Federal agencies in the region and neighboring Ontario, are currently monitoring amphibian populations; results of these efforts will provide the NPS with a broader context for data collected in parks of the GLKN.

1.3 Measurable Objectives

A characteristic of most amphibian species is a high reproductive potential (hundreds to thousands of eggs) and high variance in annual abundance due to local weather conditions and its effects on recruitment each year (Lannoo 2005). For example, droughts may compromise larval development, or deep freezes may reduce the number of breeding adults in a local population, but recovery can be rapid once favorable conditions return. A graph of the number of individuals across years normally has steep peaks and valleys, reflecting variation in numbers each year. With these demographic traits, tracking abundance is not only expensive (mark-recapture studies or counting of individuals for relative abundance indices is required), but not particularly informative of population sustainability for most species. Occupancy of habitats is a more feasible metric to track, requiring only a presence or absence determination during each sampling period (or some probability thereof), and the trend in occupancy over a study area is a measurable indicator of changes in population distribution and overall abundance (MacKenzie et al. 2006). We therefore use an occupancy monitoring approach.

The original program goal was to design and implement a monitoring program to track changes in the occupancy distribution of selected amphibian species over time, treating each park independently as well as pooling data regionally, and allowing for flexibility in the sampling design to address specific park species and habitats. The program aim was to maintain high standards for statistical power and percent change. The initial challenge was to develop a protocol and study design to provide 80% power to detect a 30% change in abundance over 5 years in each park. In researching

this challenge several problems arose. First, barring unlimited resources, it is not likely to be possible to achieve 80% power to detect a 30% change in abundance over only 5 years (Hayes and Steidel 1997). More likely, for this power and effect size with 0.05 significance, about 20 years would be required to detect a trend, by which time the trend and its causes may be too advanced for management responses to be effective. Power to detect a temporal trend will depend on the magnitude in year to year fluctuations in occupancy. Studies which approach these levels of power and effect use designs precluded by fiscal constraints for most projects, with many sites sampled multiple times (e.g., Manley et al. 2005; with 158 sites sampled twice). Therefore, a feasible plan required down-scaling these inspirational objectives and/or lengthening the timescale over which the objectives are to be evaluated.

Ultimately we were fiscally constrained to sampling 10 sites per park, and sampling necessarily involves a variety of wetland types. The final protocol achieves near perfect (>95% detection) annual detection of most species at these sites, with the ability to model how covariates such as habitat type affect occupancy trends, and providing very accurate annual occupancy estimates at the site level which will be useful for management. For species with near perfect detection direct use of data to examine occupancy is feasible (calculating occupancy estimates adds no new information). Pooling data across parks for modeling regional trends will also be possible for those species occupying more than 30 sites. Data collected to date (2013-2017) show that this will be feasible for several widespread species within the dataset for the seven parks (e.g., several species occupy over 40 monitored sites across parks). We also recommend that other parties in the region be encouraged to implement the protocol to increase the number of sites monitored and pool data to improve regional modeling (i.e., other federal and state agencies and NGOs with interest in long term ecological monitoring), especially to allow for modeling on smaller spatial scales (e.g., Lake Michigan Basin only). Moreover, during the initial data synthesis (Casper et al. 2018) we developed a method to utilize call index values as a measure of how population size fluctuates annually, to inform site level assessment. While not part of this protocol, the network has adjusted data analysis to collect a call index value metric on a pilot basis.

It is clear that methods and models for monitoring amphibians are changing rapidly (Marsh and Trenham 2008; Hui et al. 2011; Liu et al. 2011; Miller et al. 2011, 2013; Pillay et al. 2014). Therefore our intention in this protocol is to adequately track detection probabilities (DP), and collect and archive data that can be used in existing and future models. Incorporating DPs into modeling is essential, therefore study design must allow for the ability to calculate DPs from the field data. Insufficient sampling is the most common shortcoming of existing programs, resulting in wide variance in confidence intervals for occupancy estimates. DPs are known to be heterogeneous, varying both spatially and temporally, so borrowing DPs from other studies is not recommended. Instead, investigators should: a) plan to sample a subset of sites within each study area to calculate local DPs for each sampling period (i.e., annually or single season), or b) only use species/method combinations where DPs are known to be both high and have little variation under protocols constraining all confounding variables such as temperature (i.e., acoustic Spring Peeper surveys) – such that the DP can be ignored or at least considered constant. Mathematically, the minimum number of replicates for calculating an average DP is three, but significantly more replicates are

recommended because the DP is inherent to base calculations feeding into models and if inaccurate will make results misleading and non-representative of actual conditions (Johnson 2002). The relationships between DP and the minimum number of samples needed for detecting a species if it is present at a given confidence level, and the inverse – the percent confidence in detecting a species if it is present given a set number of samples – is illustrated in Table 1.

Table 1. Number of samples and confidence achieved for a given detection probability (DP).*

	N Samples										Minimum Number of Samples Needed to Achieve:		
Average DP	**1**	**2**	**3**	**4**	**5**	**6**	**7**	**8**	**9**	**10**	**75% Conf.**	**85% Conf.**	**95% Conf.**
0.10	10%	19%	27%	34%	41%	47%	52%	57%	61%	65%	14	19	29
0.15	15%	28%	39%	48%	56%	62%	68%	73%	77%	80%	9	12	19
0.20	20%	36%	49%	59%	67%	74%	79%	83%	87%	89%	7	9	14
0.25	25%	44%	58%	68%	76%	82%	87%	90%	92%	94%	5	7	11
0.30	30%	51%	66%	76%	83%	88%	92%	94%	96%	97%	4	6	9
0.35	35%	58%	73%	82%	88%	92%	95%	97%	98%	99%	4	5	7
0.40	40%	64%	78%	87%	92%	95%	97%	98%	99%	99%	3	4	6
0.45	45%	70%	83%	91%	95%	97%	98%	99%	100%	100%	3	4	6
0.50	50%	75%	88%	94%	97%	98%	99%	100%	100%	100%	2	3	5
0.55	55%	80%	91%	96%	98%	99%	100%	100%	100%	100%	2	3	4
0.60	60%	84%	94%	97%	99%	100%	100%	100%	100%	100%	2	3	4
0.65	65%	88%	96%	98%	99%	100%	100%	100%	100%	100%	2	2	3
0.70	70%	91%	97%	99%	100%	100%	100%	100%	100%	100%	2	2	3
0.75	75%	94%	98%	100%	100%	100%	100%	100%	100%	100%	1	2	3
0.80	80%	96%	99%	100%	100%	100%	100%	100%	100%	100%	1	2	2
0.85	85%	98%	100%	100%	100%	100%	100%	100%	100%	100%	1	1	2
0.90	90%	99%	100%	100%	100%	100%	100%	100%	100%	100%	1	1	2
0.95	95%	100%	100%	100%	100%	100%	100%	100%	100%	100%	1	1	1

*Survey effort above double line (confidence <0.3) is not recommended, and is insufficient even for occupancy modeling. Survey effort below the single line achieves 95% confidence with no need to correct for false absences. Minimum number of samples needed = LOG(significance level)/LOG(1-DP). Percent confidence in detecting species = 1-(1-DP)^(N samples).

Prior work has shown that acoustic frog surveys, with sufficient replicates, achieve sufficiently high and consistent DPs for occupancy modeling of most species in the western Great Lakes (Hecnar and Casper 2009). These surveys can be automated with ARS (i.e., Song Meters) to make them more economical. We have tested Song Meters in the field for six seasons and found them to be reliable in collecting audio recordings, and this technology is rapidly expanding in use. A number of visual search techniques also show great promise for cheaply monitoring additional ecologically important amphibian species, with several SOPs available (Hecnar and Casper 2009). In particular, terrestrial searches for the Eastern Red-backed Salamander, and aquatic/shoreline searches for eggs of Wood Frog, Four-toed Salamander and ambystomid salamanders *(Ambystoma laterale, A. maculatum, A. tigrinum, A. opacum*), return consistently high DPs with three 30-minute searches, and are also recommended for long-term monitoring in the western Great Lakes as a *Supplemental* protocol where staffing is available (including through partner programs). There is increasing evidence that monitoring salamanders may be more ecologically meaningful than monitoring frogs, owing to their comparatively poor mobility, underscoring the importance of this potential supplemental monitoring (Harper et al. 2008). If future results warrant, the program (or individual parks) may even consider shifting the primary focus to salamanders altogether.

The *Core* protocol will allow for scaling through increased sampling effort as future funding may allow. The initial effort will deploy ARS at ten permanent sites per park in a repeated measures design (commensurate with the ARMI TWGCRN initiative), potentially adding additional sites in the future if funding and the additional work load is feasible (30 sites is the target goal for regional trend analyses, which can be achieved within a park, or by pooling sites across parks or with other participating parties in a region). This provides opportunity for shortening the time required to detect change if funds and staff are available.

Data from simultaneous monitoring of environmental conditions, including potential stressors, will be used to examine whether any trends in amphibian populations are associated with trends in environmental parameters. Data from monitoring amphibians will also be integrated with data from monitoring other vital signs to assess the long-term change in ecological conditions in the GLKN. The amphibian monitoring program is intended to be a part of a larger environmental monitoring effort.

1.3.1 Specific objectives:

1. Annually sample for the presence of amphibians at ten permanent locations in each park, using automated acoustic survey methods which maximize target species detection. Sampling may be supplemented by other methods subject to approved standard operating procedures as they become available.
2. Periodically assess the percent area occupied (PAO) over time to determine if the distribution or number of occupied sites of any monitored species changes significantly and compare results with results from other national and state programs. The PAO (MacKenzie et al. 2002, MacKenzie et al. 2003) will be the principal metric to quantify occupancy of sites for targeted species across parks, and requires analyzing data for each species independently for each sampling method used.

3. Where indicated by significant trends in PAO, use available measures of environmental conditions to test whether trends in occupancy are associated with environmental parameters (i.e., as covariates for PAO analyses). For example, precipitation, drought index, habitat fragmentation, temperature, ice duration, snow cover, frost depth, water chemistry, etc. Some of these measures will be obtained during sampling for amphibians, and others may be available from other GLKN or partner monitoring programs (e.g., Elias et al. 2008, Magdalene et al. 2008, Kennedy et al. 2010; various weather data sources).

2.0 Sampling Design

2.1 Rationale

We considered and tested the efficiency and effectiveness of several standard survey methods for amphibians in our prior studies (Bowen and Beever 2010, Hecnar and Casper 2009), and reviewed relevant literature (Hayes and Steidel 1997, Bridges and Dorcas 2000, Manel et al. 2001, Buech and Egeland 2002, Dodd 2003, Paton et al. 2003, Manley et al. 2005, Weir et al. 2005, MacKenzie et al. 2006, Bailey et al. 2007a-b, Marsh and Trenham 2008, Waddle et al. 2009, Weir et al. 2009, Hui et al. 2011, Liu et al. 2011). We first discarded sampling methods for species-method combinations which yielded detection probabilities too low to be useful for occupancy modeling (<0.3; MacKenzie and Royle 2005). We then took into account the effort (manpower and other resources) required for conducting the sampling, potential logistical difficulties, and the cost of the different methods. We also considered the biology of the amphibian species detected by each method and how trends in these species may be ecologically informative in the context of the overall goals of the GLKN's monitoring program; aiming towards choosing both long-lived, low fecundity species, and short-lived, high fecundity species (Harper et al. 2008). Finally, we recognize that sampling methods for specific amphibian species are under continuing development and refinement, and designed a modular protocol which allows for adding additional methods as these become available. For example, in the GLKN region, ongoing work on sampling methods for the Common Mudpuppy and the Four-toed Salamander are of particular interest, given the ecological requirements for these species (aquatic benthic and mature mesic hardwood forest associates, respectively); as are refinement of methods for monitoring pond breeding salamanders fitting the long-lived, low fecundity model (i.e., visual searches for eggs and adults, aquatic funnel traps).

Daytime visual or trapping surveys can be used to monitor all amphibian species in the GLKN (Heyer et al. 1994). The same is not true for nighttime acoustic surveys because all species within the GLKN do not call during breeding (Tables 2A, 2B; Conant and Collins 1998). Data collected through any sampling method can be analyzed for PAO if surveys are properly designed, but different methods require different resources and expertise, sample different species suites, and can result in important differences in what is known about populations. We outline some of these differences in Table 3. Comparisons 1, 2, 5, and 6 favor daytime visual or trapping surveys, whereas comparisons 3, 4, 7, 8, and 9 favor nighttime acoustic surveys. Based upon these comparisons and the lessons learned from our pilot work, we chose nighttime acoustic surveys as the primary preferred method to monitor amphibian occupancy, patterned after the NAAMP protocol (Weir and Mossman 2005). We make this choice assuming that surveys will be conducted using ARS to overcome sample size issues, logistic and human resource limitations, and reduce program costs.

Table 2A. Occurrence and characteristics of 23 frog and toad species in GLKN parks. APIS= Apostle Islands National Lakeshore, GRPO= Grand Portage National Monument, INDU= Indiana Dunes National Lakeshore, ISRO= Isle Royale National Park, MISS= Mississippi National River and Recreation Area, PIRO= Pictured Rocks National Lakeshore, SACN= St. Croix National Scenic Riverway, SLBE= Sleeping Bear Dunes National Lakeshore, VOYA= Voyageurs National Park.

Species (Common Name)	Parks where confirmed or likely present									Terrestrial Phase?	Life History[1]	Type of Wetland[2]	Breeding calls?	Breeding Phenology[3]
	APIS	GRPO	INDU	ISRO	MISS	PIRO	SACN	SLBE	VOYA					
Blanchard's Cricket Frog	–	–	X	–	X	–	–	–	–	Y	<1 / <400	P, S	Y	L
Spring Peeper	X	X	X	X	X	X	X	X	X	Y	2 / 700–1,000	S, T	Y	E
Boreal Chorus Frog	X	X	–	X	X	?	X	–	X	Y	1–3 / 500–1,459	S, T	Y	E
Western Chorus Frog	–	–	X	–	–	?	–	X	–	Y	1–3 / 500–1,459	S, T	Y	E
Cope's Gray Treefrog / Gray Treefrog	X	X	X	X	X	X	X	X	X	Y	7 / ca. 1,800	S, T	Y	M
Eastern American Toad	X	X	X	X	X	X	X	X	X	Y	5 / 2,000–20,000	P, S, T	Y	E-M
Fowler's Toad	–	–	X	–	–	–	–	X	–	Y	5 / 2,000–10,000	P, S, T	Y	E-M
American Bullfrog	X	–	X	–	?	?	X	X	–	Y	10 / up to 20,000	P	Y	L
Green Frog	X	X	X	X	X	X	X	X	X	Y	5 / 1,000–7,000	P	Y	L
Pickerel Frog	–	–	–	–	–	–	X	X	–	Y	X / X	P, S	Y	E-M

[1] Estimated longevity in years / clutch size range (from Lannoo 2005); X = unknown.

[2] P = permanent (inc. lakes and streams), S = semi-permanent (fish absent), T = temporary.

[3] Spawning timing: E = early, M = mid-season, L = late.

Table 2A (continued). Occurrence and characteristics of 23 frog and toad species in GLKN parks. APIS= Apostle Islands National Lakeshore, GRPO= Grand Portage National Monument, INDU= Indiana Dunes National Lakeshore, ISRO= Isle Royale National Park, MISS= Mississippi National River and Recreation Area, PIRO= Pictured Rocks National Lakeshore, SACN= St. Croix National Scenic Riverway, SLBE= Sleeping Bear Dunes National Lakeshore, VOYA= Voyageurs National Park.

Species (Common Name)	Parks where confirmed or likely present									Terrestrial Phase?	Life History[1]	Type of Wetland[2]	Breeding calls?	Breeding Phenology[3]
	APIS	GRPO	INDU	ISRO	MISS	PIRO	SACN	SLBE	VOYA					
Northern Leopard Frog	X	X	X	–	X	X	X	X	X	Y	5 / 645–7,648	P, S	Y	E-M
Mink Frog	X	X	–	X	–	X	X	–	X	Y	6 / 500–4,000	P	Y	L
Wood Frog	X	X	X	X	X	X	X	X	X	Y	3–5 / 300–1,500	S, T	Y	E

[1] Estimated longevity in years / clutch size range (from Lannoo 2005); X = unknown.

[2] P = permanent (inc. lakes and streams), S = semi-permanent (fish absent), T = temporary.

[3] Spawning timing: E = early, M = mid-season, L = late.

Table 2B. Occurrence and characteristics of 23 salamander species in GLKN parks. APIS= Apostle Islands National Lakeshore, GRPO= Grand Portage National Monument, INDU= Indiana Dunes National Lakeshore, ISRO= Isle Royale National Park, MISS= Mississippi National River and Recreation Area, PIRO= Pictured Rocks National Lakeshore, SACN= St. Croix National Scenic Riverway, SLBE= Sleeping Bear Dunes National Lakeshore, VOYA= Voyageurs National Park.

Species Common Name	Parks where confirmed or likely present									Terrestrial Phase?	Life History[1]	Type of Wetland[2]	Breeding calls?	Breeding Phenology[3]
	APIS	GRPO	INDU	ISRO	MISS	PIRO	SACN	SLBE	VOYA					
Mudpuppy	X	X	X	X	X	X	X	X	–	N	32 / 36–85	P	N	E-M
Eastern Newt	X	X	X	X	X	X	X	X	X	Y	15 / 200–375	P, S	N	M
Eastern Red-backed Salamander	X	X	X	–	–	X	X	X	–	Y	X / 1–14	Terrestrial	N	M-L
Northern Slimy Salamander	–	–	X	–	–	–	–	–	–	Y	>5 / 13–34	Terrestrial	N	M-L
Blue-spotted Salamander	X	X	X	X	X	X	X	X	X	Y	X / 196–250	S, T	N	E
Spotted Salamander	X	–	X	–	–	X	X	X	–	Y	20–30 / 4–1000	S, T	N	E
Marbled Salamander	–	–	X	–	–	–	–	–	–	Y	8–10 / 30–200	T	N	Autumn
Eastern Tiger Salamander	–	–	X	–	X	–	X	X	–	Y	16 / 5–122	S, T	N	E
Four-toed Salamander	X	–	X	–	–	X	X	X	–	Y	9 / 4–80	S, T	N	M
Lesser Siren	–	–	X	–	–	–	–	–	–	N	7 / 200–500	P, S	N	E-M

[1] Estimated longevity in years / clutch size range (from Lannoo 2005); X = unknown.

[2] P = permanent (inc. lakes and streams), S = semi-permanent (fish absent), T = temporary.

[3] Spawning timing: E = early, M = mid-season, L = late.

Table 3. Comparisons between daytime and nighttime occupancy survey methods. Bold and underlined numbers indicate the more advantageous cell in the comparison of the two cells across that row.

Daytime Visual, Trapping and Call Surveys	Nighttime Call Surveys
1. Can detect all life stages of all potential target species, including species that do not call.*	1. Can detect only adults of potential target species that call.
2. Requires repeat surveys during the time when individuals of any targeted life stage (embryos, larvae, metamorphs, sub-adults, adults) are present. This is often a longer detection time window than for nighttime surveys.*	2. Requires repeat surveys during the time when calling males are present. This is often a shorter detection time window than for daytime surveys.
3. Fewer potential breeding sites/replicates can be sampled per person-hour (typically 30-minutes per survey).	**3**. More potential breeding sites/replicates can be sampled per person-hour (typically 10-minutes per survey). ARS can be used to further reduce human effort.*
4. Identification of some amphibian species in the GLKN, especially larvae, can be difficult or indeterminate.	**4**. Identification of calling male anuran species in the GLKN is relatively easy.*
5. Daytime surveys allow for a larger area of inference because boating and walking to sites can be done safely during the day.*	5. Nighttime call surveys restrict areas of inference to smaller areas than for daytime surveys because call surveys cannot be done safely and efficiently at night in areas without roads that can be traversed easily. However, use of ARS set up during the daytime can eliminate this difference.
6. Can be conducted safely in all parks of the GLKN.*	6. Cannot be conducted safely in all park areas unless ARS are used.
7. Requires significant training and experience for surveyors to identify egg masses and larvae and to conduct surveys correctly. Also requires that crews are physically fit.	**7**. Requires relatively less experience for surveyors to learn calls of species in each park and to identify them successfully. Physical fitness not as important with fewer sites sampled by ARS.*
8. Depending upon the park and the sampling design, daytime surveys require two people, one vehicle, one or two types of boats, bushwhacking, possibly occasional transport by float plane, boat safety training, aircraft safety training, and substantial first-aid training.	**8**. Logistics not as onerous with fewer sites sampled by ARS.*
9. Results from daytime surveys are relatively less comparable with other monitoring programs in the region because fewer programs conduct daytime surveys.	**9**. Results from nighttime call surveys are more comparable with those from other regional monitoring programs because the majority of them conduct nighttime call surveys.*

The need for survey replication at sampled sites to estimate detection probabilities (DP) creates a trade-off between the number of sites sampled and the number of repeated surveys at each site. For multi-season trend modeling, research suggests that a minimum of 30 sites should be sampled, with each site sampled multiple times for calculating DPs. From MacKenzie et al. (2006) we learn that a standard design, where multiple sites are surveyed multiple times each season, is the most robust and

allows for the most modeling options. All other designs suffer from assumptions that are known to be frequently violated, such as assumptions that DPs do not vary among years (i.e., double sampling design, removal design). For multi-season occupancy trend analyses, unconditional models allow for not all sites being sampled each year, because of the ability to incorporate missing values (but a reasonable fraction should be surveyed each year), and the implicit dynamics model can be used where the same or different sites are surveyed each year. Bailey et al. (2007b) has examined the need for temporal replication at sampled sites to estimate detection probability and the trade-off this creates between spatial replication (number of sample sites) and temporal replication (number of repeated surveys at each site). In general, the standard design outperforms the double-sampling design, and the removal design is only recommended if detection probability is constant across surveys (it almost never is).

Occupancy modeling is a rapidly changing field, and new or adjusted modeling techniques are expected to continue evolving. Differing approaches and healthy debate are evident among ecologists and statisticians (Manel et al. 2001, Marsh and Trenham 2008, Hui et al. 2011, Liu et al. 2011). This argues for a robust, standard sampling design, which allows for the most flexibility in future analyses.

Under these constraints, we chose to establish a Core monitoring protocol using calling surveys with ARS at a minimum of ten permanent sites per park (five for small parks), and add more sites over time as funding allows. Second, we provide a Supplemental SOP for time constrained visual searches for additional species if resources allow. Initial ARS deployment (65 units) will be: ten per park at INDU, SLBE, PIRO, ISRO, APIS, and MISS, and five at GPRO. Each unit will include external temperature sensors to monitor air (and optionally water) temperature. Photo documentation of water level is also recorded on each site visit, and water level meters may be considered as a future additional option.

Additional amphibian research and monitoring is conducted at VOYA under the TWGCRN. VOYA staff deploy ten ARS units (provided by GLKN) and data may be available to both GLKN and TWGCRN to analyze independently. SACN is already an established research node for the TWGCRN, with amphibian monitoring initiated in 2008. At SACN, USGS owns the equipment, conducts all the fieldwork, and does all the analysis. The TWGCRN approach is still under development at the time of this writing, but involves the use of satellite and airborne sensors, as well as temperature sensors on the ground deployed immediately adjacent to wetlands. The TWGCRN network is based upon the use of ARS units with a focus on amphibians, but also including other elements of the soundscape, ranging from birds to insects to humans. The TWGCRN protocol is expected to record five minute samples on the top of every hour, from before ice-out to late August or into September, collecting voluminous data. The GLKN *Core* protocol will collect data in a similar fashion, identical to a subset of the data collected by the ARMI TWGCRN, allowing for direct collaboration and comparisons. See SOP #5 (Deploying and Retrieving Automated Recording Systems) for details on ARS deployment.

The target populations for the GLKN *Core* protocol study design are those anuran species which achieve robust enough sampling for occupancy modeling (>30 sites across a region), with occupancy

evaluated ca. every 5 years. Rather than make *a priori* assumptions about habitat suitability for species' occurrence (such as based on wetland type), the number of monitored sites occupied by each species will be adjusted annually with species' occurrence in any year justifying the site inclusion in the model. This design is warranted given the near perfect detection rates for occupancy achieved. It also eliminates the inclusion of unsuitable sites in the models which could bias trend estimates, favoring detection of declines as opposed to increases, at least in the early years. This is acceptable as the main program objective is to detect declines. Additionally, all monitored sites will have a near perfect record of site occupancy for individual site assessment and management.

2.1.1 Target species

Amphibians in the GLKN are mostly generalist species in terms of habitat requirements because the systems have only had a few thousand years to evolve since the last glaciation (although species may be evolving towards finer niche specialization), and for many species ranges continue to expand or contract with climate change. The selection of target species is closely integrated with monitoring methods, site selection, program feasibility, and detection probability. For the *Core* protocol using acoustic surveys, target species are the 14 species of anurans found in the network: Blanchard's Cricket Frog (*Acris blanchardi*), American toad (*Anaxyrus americanus*), Fowler's Toad (*Anaxyrus fowleri*), Cope's Gray Treefrog (*Hyla chrysoscelis*), Gray Treefrog (*Hyla versicolor*), Spring Peeper (*Pseudacris crucifer*), Boreal Chorus Frog (*Pseudacris maculata*), Western Chorus Frog (*Pseudacris triseriata*), American Bullfrog (*Lithobates catesbeianus*), Green Frog (*Lithobates clamitans*), Northern Leopard Frog (*Lithobates pipiens*), Pickerel Frog (*Lithobates palustris*), Mink Frog (*Lithobates septentrionalis*), and Wood Frog (*Lithobates sylvaticus*). A variety of species specific filters are used in data analyses to improve DPs for occupancy modeling (for example, by constraining samples by season, time of day, and temperature), and two independent data analyses (automated and manual) are performed to achieve >95% confidence in detection for most species (see SOP #7). Important characteristics of the target species are that they breed in wetlands, call during breeding, and the calls are readily heard (Tables 2a, 2b, Figure 1). Species that have short calling windows (Wood Frog), or call relatively quietly (Northern Leopard Frog, Mink Frog, Wood Frog), are detected less reliably (but still at levels quite suitable for occupancy modeling), while amphibian species that do not call (salamanders) cannot be monitored by this method. For salamanders, a Supplemental SOP is provided, using time constrained visual searches.

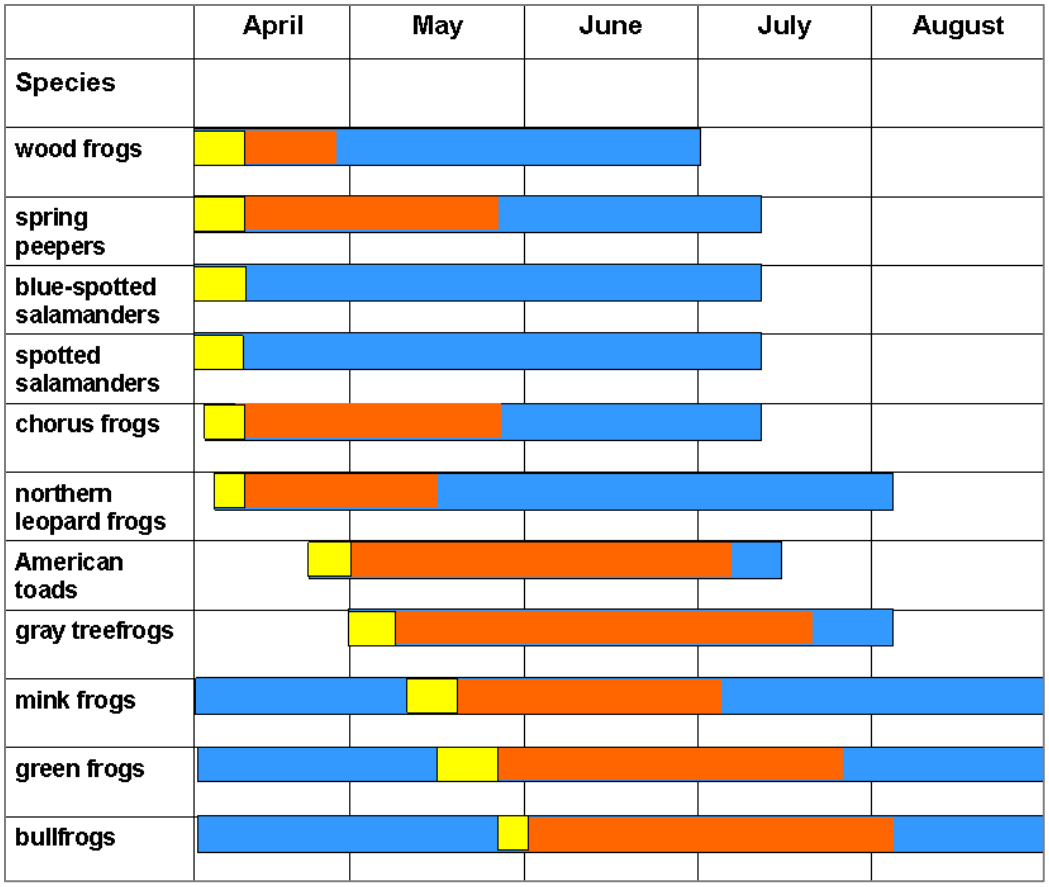


Figure 1. Typical reproduction season for several species at St. Croix National Scenic Riverway. Yellow = period when males call during warm springs. Orange = period when males call more typically. Blue = total reproduction season, including larval period.

The target species recommended for the initial *Core* protocol represent three breeding habitat classes (Tables 2a, 2b): temporary to semi-permanent wetlands (Spring Peeper, both Chorus Frogs, Wood Frog), semi-permanent to permanent wetlands (Blanchard's Cricket Frog, both toads, both treefrogs, Northern Leopard Frog, Pickerel Frog), and permanent wetlands (American Bullfrog, Green Frog, Mink Frog). This habitat stratification is taken into account in the study design. Another factor in selecting target species is ecological context (Table 4). This includes factors such as rarity and range limits which might make the species more sensitive to climate or other change, hence a more appropriate "indicator species." Local rarity and status will be assessed through each park. Of the target species selected, eight are at or near range limits within the network (Table 4; Blanchard's Cricket Frog, Fowler's Toad, Cope's Gray Treefrog, Gray Treefrog, American Bullfrog, Pickerel Frog, Boreal Chorus Frog, Western Chorus Frog). Monitoring of seven salamander species with range limits within the network may also be important, but will require different methods: Spotted Salamander (*Ambystoma maculatum*), Marbled Salamander (*Ambystoma opacum*), Eastern Tiger Salamander (*Ambystoma tigrinum*), Four-toed Salamander (*Hemidactylium scutatum*), Eastern Red-backed Salamander (*Plethodon cinereus*), Northern Slimy Salamander (*Plethodon glutinosus*), and Lesser Siren (*Siren intermedia*).

Table 4. Target species characteristics. Range Limit refers to whether or not the species range is limited within the network region, these species are expected to be more sensitive to regional climate change.

Species	# Parks	Range Limit	Call Surveys	Habitat Context	Best Monitoring Potential
Spotted Salamander	5	Yes	No	Hardwood forest, ephemeral wetlands	egg searches, aquatic funnel traps
Blue-spotted Salamander	9	No	No	Hardwood forest, ephemeral wetlands	egg searches, aquatic funnel traps
Marbled Salamander	1	Yes	No	Hardwood forest, ephemeral wetlands	egg searches, aquatic funnel traps
Eastern Tiger Salamander	4	Yes	No	Grasslands, savanna, ephemeral wetlands	egg searches, aquatic funnel traps
Four-toed Salamander	5	Yes	No	Hardwood forest, ephemeral wetlands	nest searches
Eastern Newt	9	No	No	Grasslands, savanna, semi-permanent wetlands	aquatic funnel traps
Eastern Red-backed Salamander	6	Yes	No	Hardwood forest	terrestrial searches
Northern Slimy Salamander	1	Yes	No	Hardwood forest	terrestrial searches
Western Lesser Siren	1	Yes	No	Aquatic, permanent to semi-permanent lakes and streams	aquatic funnel traps
Mudpuppy	8	No	No	Aquatic, permanent lakes and streams	aquatic funnel traps
Blanchard’s Cricket Frog	1	Yes	Yes	Shoreline, permanent to semi-permanent lakes and streams	call surveys, shoreline searches
Eastern American Toad	9	No	Yes	General terrestrial, permanent to semi-permanent wetlands, lakes and streams	call surveys
Fowler’s Toad	2	Yes	Yes	Sandy dunes and woodlands, permanent to semi-permanent wetlands, lakes and streams	call surveys
Cope’s Gray Treefrog	5	Yes	Yes	Sandy savannas and woodlands, temporary to semi-permanent wetlands	call surveys
Gray Treefrog	9	No	Yes	Forests and woodlands, temporary to semi-permanent wetlands	call surveys

Table 4 (continued). Target species characteristics. Range Limit refers to whether or not the species range is limited within the network region, these species are expected to be more sensitive to regional climate change.

Species	# Parks	Range Limit	Call Surveys	Habitat Context	Best Monitoring Potential
Spring Peeper	9	No	Yes	Forests and woodlands, temporary to semi-permanent wetlands	call surveys
Boreal Chorus Frog	6	Yes	Yes	Grasslands and savannas, urban settings, temporary to semi-permanent wetlands	call surveys
Western Chorus Frog	2	Yes	Yes	Grasslands and savannas, muskeg, aspen parklands, urban settings, temporary to semi-permanent wetlands	call surveys
American Bullfrog	6	Yes	Yes	Shorelines, permanent lakes and streams	call surveys, shoreline searches
Green Frog	9	No	Yes	Shorelines, semi-permanent to permanent lakes and streams	call surveys, shoreline searches
Northern Leopard Frog	8	No	Yes	Shorelines, forest, grasslands, semi-permanent to permanent lakes and streams	call surveys, shoreline searches
Pickerel Frog	2	Yes	Yes	Shorelines, forest, woodlands, semi-permanent to permanent lakes and streams, usually springs	call surveys, shoreline searches
Mink Frog	6–7	Yes	Yes	Shorelines, permanent lakes and streams	call surveys, shoreline searches
Wood Frog	9	No	Yes	Forests and woodlands, temporary to semi-permanent wetlands	egg searches, call surveys

2.1.2 Analytical Methods

During program development we collected acoustic data from seven parks and over 30 other sites within the network region to obtain training files (confirmed species recordings), including from Grant, Iron, Milwaukee, Ozaukee, Sauk, and Washington Counties in Wisconsin, and the western Upper Peninsula of Michigan. We obtained Fowler's Toad training files from Long Point, Ontario. This effort confirmed the efficacy of data collection and reliability of recording equipment, and samples were used to develop and test automated recognizers in Song Scope software (Wildlife Acoustics, Inc., 970 Sudbury Road, Concord, MA 01742-4939). This process is described in the Song Scope software manual. As of this writing we have acceptable recognizers developed for all network anuran species except Fowler's Toad and Pickerel Frog, which are under development. However, three of the recognizers do not distinguish between individual species in species pairs with very similar calls: Mink/Green frogs, Gray/Cope's Gray treefrogs, and Boreal/Western Chorus frogs. These species can be distinguished by manual scanning (acoustic and visual examination of spectrograms) and/or geography (see SOP #7 and Tables 2A, 2B).

Recognizer parameters were tuned to minimize false presence rates (at the expense of higher false absence rates), and false presence rates are further reduced by filtering sample selection prior to running recognizers, and filtering of recognizer results (detailed in SOP #7). We determined false absence and false presence rates for each recognizer by manually scanning automated detection results to determine how many were correct. Manual scanning involves visually scanning the spectrogram in Song Scope software for the call signature at the location reported by the recognizer, then playing it back acoustically for final confirmation (see SOP #7). In our test data from 2013 through 2015, 3,900 of 4,895 auto-detections examined were correct (Appendix A available as MS Excel spreadsheet). False presence rates in recognizer results ranged from 0.000 to 0.730 for individual detections, but were zero for site occupancy (that is, wherever false detections were discovered, valid detections were also always found). Therefore, we are confident that by coupling recognizer results with manual scanning to confirm at least one good detection, false presences are eliminated on an annual site occupancy basis.

Despite the persistence of false presences in the recognizer results, because of the large sample sizes obtained (100s) the probability of returning a false absence for occupancy of any site in any year is near zero. Only on rare occasions did we encounter false absences from automated analyses during manual scanning, usually due to interference from extraneous noise (including other frog species). We also encountered sites where, because of species rarity or because most of the calling period was missed due to the ARS being deployed too late in the season, false absences were reported (this problem affects only Wood Frogs, which have a short, sporadic, and very early calling period).

In order to survey for species without recognizers, and to provide a second independent survey data set for use in models developed by Miller et al. (2011, 2013; Pillay et al. 2014), which take into account false presences, we developed a manual scanning procedure to supplement the automated results (detailed in SOP #7). The manual procedure selects 30 samples for analysis from each site in each year. This sample size was chosen to exceed the 20 samples per cell recommended by Simmons et al. (2011). When both surveys are performed, we are confident that all network species will be

detected with >95% confidence, with the possible exception of the Wood Frog, which might be missed if ARS are consistently deployed too late to capture their calling season. In this case, occupancy modeling will be more informative.

The data obtained by using this protocol will be analyzed primarily via comparisons of models that Mackenzie and others developed and incorporated into the computer program PRESENCE (Mackenzie and Kendall 2002, Mackenzie et al. 2002, Mackenzie et al. 2003, Mackenzie and Royle 2005), and can also be implemented in other statistical packages such as R (www.r-project.org). As of this writing, we anticipate using models developed by Miller et al. (2011, 2013; Pillay et al. 2014), which take into account false presences, and will incorporate data from two independent surveys on the same dataset (automated vs. manual). With these methods, various models that involve different variables are compared in terms of their parsimony and fit to the occupancy data using information-theoretic criteria (Burnham and Anderson 2002). The percent of area occupied (PAO) is a measure of the percent of the known area of inference that is occupied by a monitored species based upon survey data.

While it may seem unnecessary to model data that achieves >95% confidence in occupancy detection, there are three primary benefits to using such models. First, they allow for estimating detection probabilities based upon repeated sampling and therefore provide a theoretically unbiased estimate of occupancy (empirically re-confirming the high confidence assumption in each analysis). In addition, measures of environmental variables can be used as covariates in the models. This latter feature enables the analyst to investigate the influence of environmental variables on the occupancy and detectability of species monitored. Finally, these models are used as the primary analytical tool for data collected by ARMI throughout the United States (Muths et al. 2006) and have been adopted by other NPS monitoring networks (Bailey et al. 2007a, Amphibian Monitoring Working Group 2008). Thus, the GLKN will be able to compare parameter estimates with those from other amphibian populations.

Each park will be sampled independently, with no assumptions of posterior pooling in analyses, although posterior pooling is recommended for post-hoc analyses to investigate regional trends. Local factors affecting amphibian decline are likely to be at least as important as regional or global factors, so the ability to perform local, per-park, analyses is important. For more information on analytical methods see SOP #7.

2.2 Site Selection and Documentation

Amphibian distribution in the GLKN is affected by two coarse habitat parameters: the breeding wetland hydro-period (for those species with aquatic stages), and the terrestrial habitat quality and class. The wetland hydro-period classes are: a) temporary (ephemeral or seasonal) wetlands, b) semi-permanent wetlands (which dry only in drought years), and c) permanent lake and stream habitats (Table 4). These coarse wetland classes represent a natural life history stratification of amphibians into three suites which exploit differing hydro-periods for larval development with differing competitive and predatory interactions (mostly with fish and macro-invertebrates). The selection of the ten permanent sampling sites considered which species were to be targeted in each park and their hydro-period requirements (see Table 4). We then stratified wetland site selection as equally as

possible among these hydro-period classes. GLKN staff assisted parks in selecting sites within the area of inference for each park using wetland, vegetation and soil layers, augmented with aerial imagery, park-staff knowledge, and ground visits. Each park chose sampling sites that best represented their habitats, target species, and wetland types. The wetland hydro-period classes will be used as a covariate in data analyses. The types of wetlands monitored may be different among network parks, depending on availability and on park-specific monitoring needs, limitations, and objectives.

In addition to wetland hydro-period, another variable that is expected to affect amphibian species occurrence is coarse terrestrial habitat class – referred to as "habitat context" in Table 4. These habitat classes will also be considered as covariates, as appropriate for each park—for example, grassland vs. forest, or deciduous vs. coniferous forest. As the program expands, additional sampling sites may be selected within under-represented terrestrial habitat classes as well.

The initial site selection identified as many potential sampling sites of each class combination as possible, in order to, a) leave room for discarding sites that fail ground truthing or must be discarded for other reasons, and b) leave room for randomized selection of additional sites if the program expands. Site selection then followed a generalized random-tessellation stratification design (Stevens and Olsen 2004) to be as spatially balanced as possible while retaining an element of randomness. Sampling sites chosen were at least 1 km apart to assure independence (the same calls not being recorded by more than one unit).

It is important to note that the stratification of sampling sites on habitat parameters that are considered suitable for the target species may set up an initial bias in the study design, such that sites are selected that are expected to be occupied, and therefore any trend detected is likely to be negative. However, detecting declines is considered to be more important than detecting increases for management purposes, hence this is deemed an acceptable bias that affords a more feasible study design. Should habitat conditions change in the future, the stratification of the sampling design will also change, and future managers should take these changes into account in expanding sampling sites and in data analyses.

The sampling area for call surveys is limited to a buffer around sampling points (the ARS unit or human listener) equal to the maximum distance at which the species can effectively be detected. This distance varies by a range of factors including call amplitude, call frequency, the relative positions of the caller and receiver, habitat, background noise (including other frogs), and the auditory acuity of the receiver (Marten and Marler 1977, Parris 2002, Parris et al. 2009). Few data are available, however, for quantifying the effective detection distance. For a conservative estimate, we recommend maintaining 1 km separation between sampling sites at this time.

The area of inference for regional modeling is all wetlands potentially occupied throughout the region modeled (sample size and region modeled will be driven by all sites ever occupied by the species modeled over time). The area of inference for the site occupancy is the site itself. Sampling site selection may be constrained by logistics within parks, such that some regions may be unavailable to survey due to the time and effort needed to reach them. However, by using ARS, such

logistic limitations should be lessened, since these sites need be visited only three times each season during daytime (once to set, once to check, and once to recover the unit annually). Surveys conducted only along roads are inherently biased because: a) the roads are not randomly located (i.e., they are often routed around the wetland habitats preferred by many amphibians); b) road-associated stressors (e.g., road salts, noise and dust generated by vehicular traffic, discarded trash, vectors of non-native species) disproportionately affect wetlands at different distances from roads; and c) the road geometry creates unequal probabilities of including different sites (e.g., a site might be accessible from portions of two different roads). Monitoring data will directly track only the area of inference, on two scales.

Final site selection included ground truthing with habitat documentation, ARS, and logger placement and mounting options were determined, GPS coordinates were taken, and a site map with directions was produced. Details of site selection and documentation are provided in SOP #3. Some parks also had priority sites to monitor such as wetlands supporting rare species. Such sites are non-random selections and do not exceed 10% of the total sites selected.

Steps Followed for Site Selection and Documentation (See SOP #3 for details)

1. Habitats, target species, and wetland types were assessed and listed for the park.
2. Target species were grouped by hydro-period and habitat requirements (Tables 2A, 2B and 4).
3. Potential sampling areas were determined considering any area of inference constraints, such as park-specific monitoring needs, limitations, and objectives.
4. GIS and other habitat data were used to identify and stratify potential wetlands within the sampling area.
5. Ten permanent sampling sites were selected from the potential wetlands identified following a generalized random-tessellation stratification design, representing the three possible wetland hydro-period classes as equally as possible.
6. ARS placement in the wetland was determined with consideration for being accessible, within a suitable habitat area, and accounting for appropriate sensor deployment. An amphibian expert (GSC) visited each site to ensure that suitable amphibian habitat was selected.
7. Site selections were ground truthed, and documentation of the exact placement and mounting method for each ARS and sensor produced.

2.2.1 Song Meter and Sensor Placement and Mounting

Existing calling survey protocols do not require users to record water temperature (i.e., NAAMP, state programs). However, they do dictate minimum water temperatures as guidelines for establishing the survey dates. Water temperatures are desirable to monitor as potentially informative covariates for data analysis. However, water temperatures can vary substantially within small distances based on temperature layering in the water column, mixing by wind, and solar radiation exposure. The ecologically relevant water temperature for frog breeding activity is the surface layer in which the adults are breeding, typically the upper few inches of the water column in shallow water zones with

emergent vegetation. We experimented with placing temperature sensors on floats to maintain the sensor at an approximately 5 cm depth, but ultimately found this to be impractical. The final protocol implements recording air temperature at the ARS location, using a solar shield. Setting water temperature loggers is optional.

Hydroperiod (whether or not ponds dry up during the sampling period) is also important to monitor. Ideally, data collected when ponds are dry should be discarded from analyses as the species is no longer present to detect. We experimented with tracking hydroperiods by deploying a temperature logger at the deepest point of ephemeral and semi-permanent ponds, and examining the seasonal log for temperature spikes characteristic of air exposure (with pond drying assumed if the sensor becomes exposed to air), but ultimately abandoned this method as too complex and prone to equipment failure for network-wide implementation. The protocol does however require photo and written documentation which can be examined to potentially discover dry periods which could be excluded from analyses. Ideally, samples taken when ponds are dry should be considered invalid and excluded from analyses, but without continuous visual monitoring this is not possible. While ignoring this causes DPs to be lower during the sampling period if in fact the pond does dry for some length of time, this is treated as an uncontrolled covariate which results in variation in DP. Species are still detected during the periods they are calling with high confidence (see SOP #7).

Ideally all sampling sites would monitor surface water temperature and hydroperiod, and at this time collection of these data at some sites is recommended to allow for a better understanding of how these covariates affect occupancy determination.

Optional: Sensor placement in permanent water bodies

For permanent water bodies if surface water temperature is to be tracked the sensor placement should be within the emergent vegetation zone or other area suitable for breeding activity, out of boat traffic lanes, and as close to the ARS mounting site as possible. The sensor should be deployed on a weighted float that will maintain it positioned approximately 5 cm below the water surface under conditions of fluctuating water levels (see SOP 3 Sec. 3.4.2). Follow the Temperature Sensor Decision Tree to determine if corded or cordless sensors should be used.

Optional: Sensor placement in ephemeral and semi-permanent ponds

The monitoring objective here is to track surface water temperature and/or hydroperiod. To monitor both, position an apparatus at the deepest point of the pond (as described in SOP 3 Sec. 3.4.2). This point should be determined during site establishment in the pilot year. To monitor hydroperiod alone, the sensor may simply be anchored at the deepest point of the pond on the pond bottom.

Temperature Sensor Decision Tree

A. **Permanent Water Bodies** (Monitoring Objective is surface water temperature only)
    1. Can Song Meter be positioned such that corded sensor will reach frog breeding habitat (i.e., shallow emergent zone)?
        a YES - use corded sensor on float system
        b NO - use HOBO sensor on float system

Note: If water recedes, sensor may become stranded and data will be discarded unless sensor is manually repositioned.

B. **Ephemeral and Semi-permanent Wetlands** (Monitoring Objective is surface water temperature and/or hydroperiod)

1. Can Song Meter be positioned such that corded sensor will reach deepest point of water body?

a YES (use corded sensor)

i. Monitoring Objective is surface water temperature and hydroperiod - use corded sensor on float system

ii. Monitoring Objective is hydroperiod only - use corded sensor on anchor system

b NO (use HOBO cordless sensors)

i. Monitoring Objective is surface water temperature and hydroperiod - use HOBO sensor on float system

ii. Monitoring Objective is hydroperiod only - use HOBO sensor on anchor system

Note: Float systems will need to be placed in protective mesh tubes to minimize entanglement, and checked and cleared if necessary during QA/QC visits. Anchored sensors should need no protection.

2.3 Sampling Frequency and Replication

Given the evolving nature of occupancy modeling methods and DPs, this protocol greatly over-samples using ARS, such that the sampling frequency and replication can be adjusted post hoc when recordings are analyzed. This allows for many replicates and tight detection probability confidence limits. Over time, these parameters can be more firmly estimated and sampling design adjusted if warranted. However, obtaining samples using ARS is cheap and large numbers of replicates can be stored even if not immediately used in analyses. We currently recommend using two independent survey methods on the data collected, one with typically hundreds of samples (up to 700 samples in the automated process), and the other with 30 samples (samples selected for manual processing), per season, with sample selection optimized for maximum detectability as detailed in SOP #7.

The ARS SOP (#5) details use, maintenance, setup and data management. The program files (e.g., *.SET, *.PGM, *SM4S) with the recording schedule will be provided by the network office. The program will schedule standard 5-minute recording sessions at the top of each hour from 5:00 p.m. to 3:00 a.m. daily (in local time), starting when the ARS is turned on at deployment, and ending 1 August, for up to 21 weeks of sampling (or simply allow the program to run until batteries fail or the unit is retrieved and turned off). To increase sampling during the primary detection time (after sunset), three additional samples will be programmed for 7:30 p.m., 8:30 p.m., and 9:30 p.m. at INDU and APIS, and at 8:30 p.m., 9:30 p.m. and 10:30 p.m. at MISS, GRPO, SLBE, PIRO, and ISRO. As SM2 models (recently discontinued) are replaced with newer SM4 models, sampling frequency may be increased as battery life is more efficient. New programming should retain the

schedule above, but may add to it, and/or increase the sample rate (which includes higher frequency sounds but increases file sizes). The ARS units are typically deployed shortly after ice-out where possible (ice-out dates will vary annually by park). Where travel during the ice-out period is problematic, ARS units can be deployed before ice-out with a delayed start programmed, in which case the network should be consulted for appropriate programming.

Frogs are generally thought to have their peak calling period shortly before and after full dark, when the darkness is warmest. Typically, calling then drops off as temperatures decrease after sunset (sometimes rapidly, especially under clear skies). These and other factors are accounted for in the sample selection for analysis (see SOP #7), whereby a subset of the recorded samples are analyzed to increase DPs.

Temperature is logged internally in each ARS unit and recorded in a text file. Values are reasonably close to the ambient air temperature if the unit is shaded, but can spike if the unit is exposed to full sun. This temperature log is not recommended for use as a covariate, use the independent air temperature data log instead, synchronized with recordings. The internal temperature log may be useful in documenting exposure to temperature extremes that might help explain ARS equipment failures.

2.4 Recommended Number of Sampling Sites

Our *a priori* power analyses and literature reviews suggest that at least 30 sites will be necessary to robustly examine trends in occupancy within a park unit. The relationships between power and sample size are shown in Figures 2 and 3 for one- and two-tailed tests. However, sampling 30 sites per park is not possible under existing fiscal and manpower constraints. Therefore, lower power and higher error rates (wider confidence limits) are a necessity under the initial program. The GLKN will deploy ten ARS units per park, one at each permanent sampling site, in a repeated measures design, and then potentially add more units depending on available funds and staff capacity. This will provide the benefit of the potentially stronger analyses provided by repeated measures for trends, and extend knowledge of diversity with additional sites added later. The ten permanent sites per park is also consistent with TWGCRN.

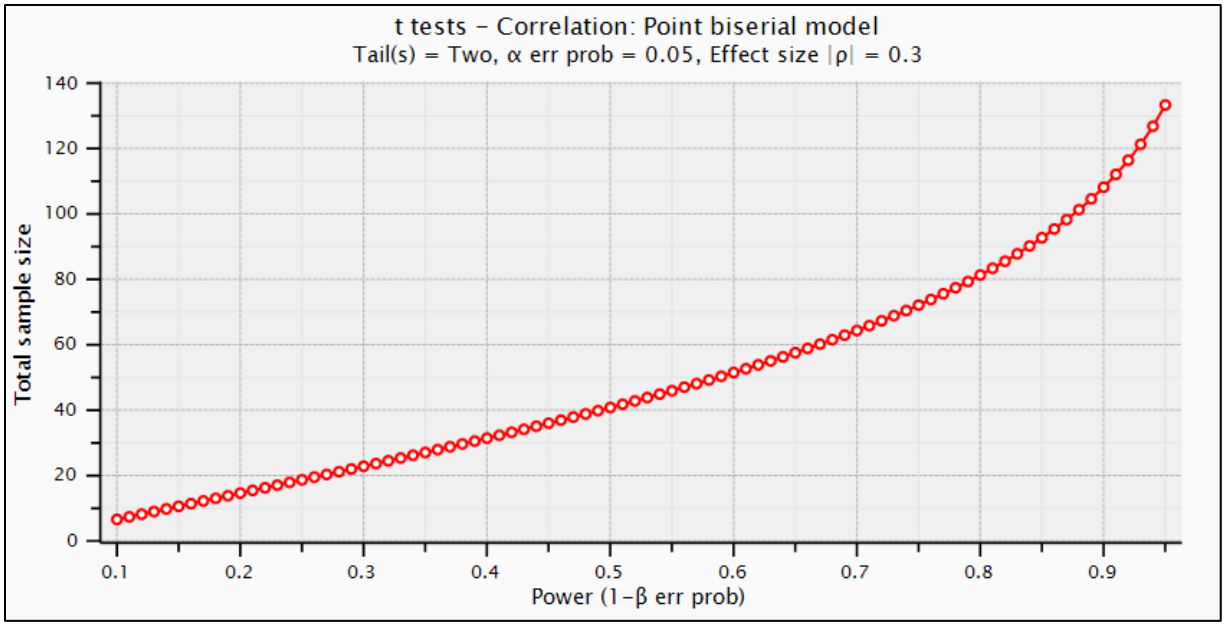


Figure 2. The relationship between power and sample size in a random design with 30% effect size and 0.05 significance for a two-tailed test.

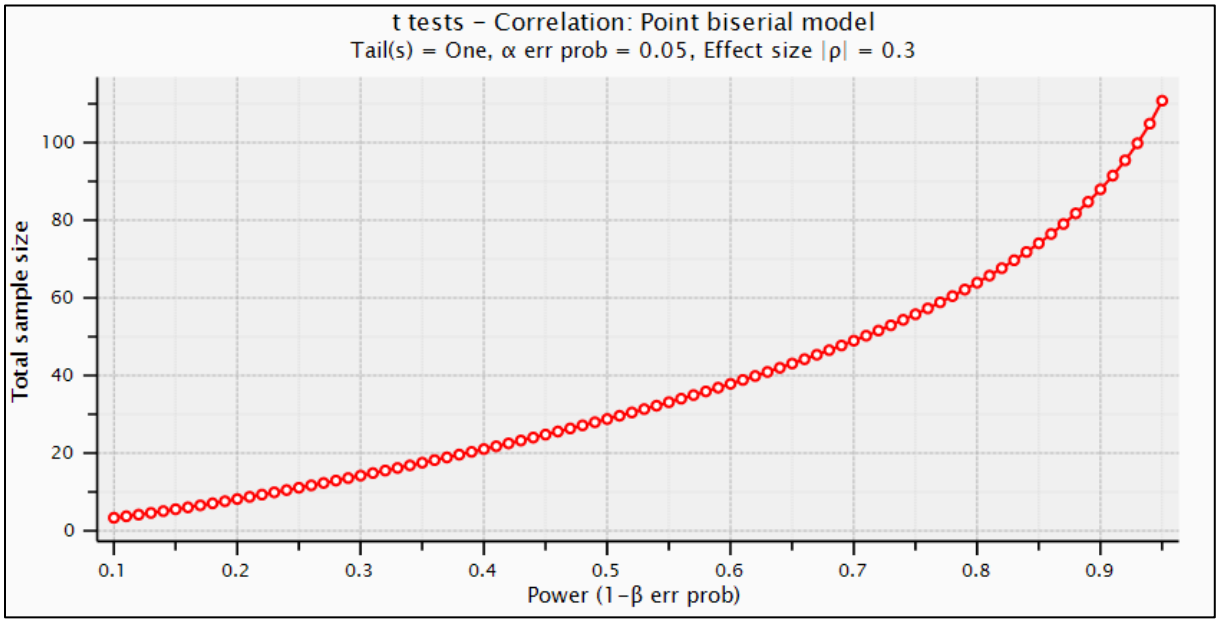


Figure 3. The relationship between power and sample size in a random design with 30% effect size and 0.05 significance for a one-tailed test.

2.5 Level of Change That Can Be Detected With This Design

The levels of change detectable (effect size) is a complex statistical issue and will vary with the number of sites successfully surveyed each season, and the number of seasons sampled for each species, as discussed in Section 1.3. We do not have results from computer simulations to describe changes detectable for each combination of parameters, and given the uncertainties involved such

simulations would probably not be very useful. However, we offer the following estimates based upon the calculations of MacKenzie (personal communication).

- If ψ (occupancy probability) = 0.5 initially, ρ (detection probability) = 0.7, 200 sites are sampled three times per season for ten seasons, and occupancy declines at a rate of 0.1, a change in ψ to 0.27 would be detectable as significant statistically based upon 95% confidence intervals. This would equate to a decline of 46% over ten years.
- If ψ = 0.8 and all other values stayed the same as above, a change in ψ to 0.6 would be significant. This would equate to a decline of 25% over ten years.
- In contrast, if ψ = 0.5 for 50 sites surveyed over ten seasons with a decline trend of 0.2 and all other values the same as above, statistical significance would occur at ψ = 0.12, a decline of 76% over ten years.
- If ψ = 0.8 for 50 sites surveyed over ten years with a decline trend of 0.2 and the same values otherwise, then significance would occur at ψ = 0.35, a decline of 56% over ten years.

The relationship between detecting levels of change and the number of sites sampled per season also demonstrates the limits of this design to detect changes in amphibian populations in each park. The above detection levels for 50 sites clearly show that very meaningful declines of populations will occur before they are detected as statistically significant based upon monitoring 10 or even 30 sites with these characteristics. Because surveying more sites per park is not realistic at this time, and the extremely high DPs achieved by this program should improve on these trend estimates, we recommend initially that a single PAO be calculated for each target species based upon sampling across all parks in which it is present per season, as well as across southern, northern, eastern and western regions (see SOP #7). This will enable monitoring the occupancy of each species over time across all occupied parks. Better results may be obtained if analyses expand pooling of data across the region through partnerships with other programs (i.e., ARMI, state programs, NGOs). These pooled statistics can be meaningful for local managers who can be alerted to changing conditions in habitat or weather regionally and how it might affect amphibians in their park.

To further arm managers with information on a local level, we also recommend that species presence for the sites monitored in each park be charted annually. Since over-sampling provides near perfect detection at each site, these estimates will provide highly accurate data on what is happening at each site, without inferring what is happening at other sites. While the accuracy of estimates of PAO within a single park for all wetlands (surveyed and not surveyed) will often be poor, occupancy estimates for the monitored sites will be near perfect, and plotting these occupancies over time will serve as an early warning system for potential declines. These data also can be complemented by other site environmental data that might provide insight into apparent declines. Managers from the GLKN can then decide to invest more resources into sampling a larger number of sites in that park, or otherwise investigate suspected change, if necessary.

2.6 Supplemental Monitoring Method

Standard Operating Procedure #11 (Visual Encounter Surveys) consists of timed visual searches for eggs or adults. These surveys result in high DPs, and 3–5 replicates of 30-minute searches during

fairly long time windows (4–10 weeks) are sufficient. Supplementing the ASR monitoring with visual searches will expand the pool of species that can be monitored to include some salamanders (which are not monitored by acoustic surveys), and increase the ability to effectively monitor Wood Frogs.

3.0 Field Methods

3.1 Preparation for the Field Season (SOP #1)

1. Applications for any required permits should be submitted far enough in advance to ensure compliance by the beginning of the field season.
2. Training and updates for field personnel should begin at least one month prior to the beginning of the field season. Make sure that all field personnel review and pass the frog call quiz for species likely to be present in the sampling region before each season begins: http://www.pwrc.usgs.gov/frogquiz/.
3. Staff should assemble, test and organize all requisite equipment beginning approximately one month prior to ice-out. This may include printing data sheets and any necessary maps, updating field folders, purchasing batteries and SD cards as needed, and checking or obtaining ARS units. Appropriate programming files should be obtained or prepared.
4. Based upon local weather conditions, staff should arrange to deploy ARS units just after ice-out, or earlier if a delayed start is programmed in. Each location should have GPS coordinates (SOP #3).

3.2 Sequence of Events During Field Season

ARS units should be visited once shortly after deployment (or shortly after start of recording if deployed with a delayed start), to ensure they are working properly. Follow the procedure in SOP #5 for checking the unit. Additional checks during the field season should be made whenever convenient, to ensure units are working properly.

3.3 Post-collection Processing of Survey Results

ARS units should be recovered at the end of the recording period (14 weeks after start of recording). Units should be cleaned, batteries removed and properly disposed of, SD cards backed up, data files stored and appropriately labeled, and data sent to the GLKN data manager. Any needed repairs or replacements should be addressed. All applicable field equipment should be inventoried, cleaned, repaired and stored in a secure location.

3.4 End-of-season Procedures

Appropriate personnel should discuss the general outcome of the field season and any changes that may need to be made to the protocol. Data management and analysis should proceed as appropriate.

4.0 Data Handling, Analyses, and Reporting

4.1 Metadata Procedures

The network's overall strategy for the generation, management, and distribution of metadata is described in chapter 8 (Data Documentation) of the GLKN Data Management Plan (Hart and Gafvert 2006) and the appendices of that document.

Executive Order 12906 mandates that federal agencies document all new geospatial data they collect or produce, either directly or indirectly using the Federal Geographic Data Committee (FGDC) Content Standard for Digital Geospatial Metadata (CSDGM; https://www.fgdc.gov/metadata/geospatial-metadata-standards). In addition, EO 12906 directs agencies to plan for legacy data documentation and provide metadata and data to the public. GLKN will also generate FGDC-style metadata for non-spatial datasets that meets this standard, absent only the geospatial-specific elements.

The FGDC Biological Data Profile (https://www.fgdc.gov/standards/projects/metadata/biometadata) contains all the elements of the CSDGM and includes additional elements for describing biological data sets. All GIS data layers must be documented with applicable FGDC metadata standards.

Metadata allows potential data users to evaluate the quality and usefulness of the data based on an understanding of the complete process under which it was collected and maintained. In this respect, all of the protocol documentation, including SOPs, are part of a dataset's metadata. A reference to the appropriate version of these documents is part of the metadata for any particular element of a dataset. The relationship between a data element and the appropriate versions of documented procedures is based on the date and time the procedures are performed. Thus the documentation must have accurate records on each version indicating the start and end date it was in effect, an indication of which version it replaced and, if applicable, the version which replaced it – both a forward and reverse documentation trail. Although perhaps obvious, all data must have an associated value for the date and time it was collected.

Most of the remaining metadata will be recorded directly in the protocol specific databases and tables. The GLKN database has fields and tables which incorporate the metadata elements specified by NPS standards. During collection of field data, these metadata elements are entered by selecting an existing profile of metadata, or by entering specific elements in data fields using pre-determined lookup values selected from dropdown menus.

4.2 Overview of Database Design

At the national level, the networks in the NPS Inventory and Monitoring Program have cooperatively developed a Microsoft Access template tool for developing monitoring databases called the NPS Natural Resource Database Template (NRDT). GLKN will implement most protocol specific monitoring databases by developing MS Access and/or ESRI personal geodatabases from the NRDT model.

The network office will maintain one master copy of the amphibian protocol-specific MS Access database on a central server. This is the only copy that can be used to export data to other locations. Additional copies (replicas) of the database can be used as a conduit for data entry and importation of data to GLKN's master version of the database. For analysis, researchers will access data from the master copy of the MS Access database, or the mirrored tables in VSIMS.

In summary, the data flow for amphibian monitoring data involves entering survey data into an Access database, and backup and storing of digital acoustic files for archive and analysis. At the end of each field season the data will receive formal QA/QC.

4.3 Data Entry, Verification and Editing

Detailed instructions for the data entry procedures for this protocol are given in SOP #6, Data Entry and Verification.

For any field data that is collected on printed data forms, and transcribed into a database later, a digital replica (scan) of the data form will be created in the MS Access database to facilitate data entry with a minimum of transcription errors.

Data verification starts with the QA/QC steps that are outlined in this protocol's Quality Assurance Plan (Gafvert and Casper 2018). As data are entered into forms of the amphibian protocol database there is a suite of QA/QC procedures that compare the entered data with expected formats and accepted data value ranges or domains. If the entered data do not pass a form-based QA/QC test the database prompts the user to make corrections and re-enter the data. For most of the form-based tests the database will not accept out-of-bound data, and correction is mandatory; for some range tests, out-of-bounds data are accepted after a user prompt, but are flagged as "suspect." QA/QC procedures on imported data (such as environmental sensor loggers) perform similar tests, but exceptions which do not pass QA/QC are presented to the user and must be reconciled before the record or dataset can be accepted by the database.

Additional comparison tests will be run on data in the master version of the MS Access database to flag records that are outside the expected range for a parameter based on previous records. These suspect data points are reviewed by an expert user and can be corrected, flagged or excluded from the dataset.

Each individual record in the amphibian monitoring dataset always has one, and only one, status flag indicating its status as provisional or final. Only after the record has passed all QA/QC steps will it be flagged as having final status. Finalized data can still be edited and changed if errors are discovered after review in reporting and analysis.

A data enterer's identification is recorded for each aspect of data handling, from collection through to the final steps of QA/QC. On data sheets, the identity of the data collector(s) is recorded and is transcribed into the appropriate data entry forms. Additionally, the MS Access database records the user name (login) for every table entry or modification. Only qualified users who have been trained and given edit permissions are allowed to edit data in the master amphibian database. These procedures protect the integrity of the data and allow the history of each data record to be traced.

4.4 Data Analyses and Reporting

As noted above, one program objective is to periodically assess the percent area occupied (PAO) over time to determine if the distribution or number of occupied sites of any monitored species changes significantly and compare these results with results from other national and state programs. The PAO (MacKenzie et al. 2002, MacKenzie et al. 2003) will be the principal metric to quantify occupancy of sites for targeted species across parks, and requires analyzing data for each species independently for each sampling method used. Available measures of environmental conditions should also be used to test whether trends in occupancy are associated with environmental parameters (i.e., as covariates for PAO analyses). For example, precipitation, drought index, habitat fragmentation, temperature, ice duration, snow cover, frost depth, water chemistry, etc. Descriptive ecological and survey data summaries will be provided as well, with metrics on species detected at each sampling site each year with detection confidence limits, detection probabilities for each species at each site, averaged detection probabilities with standard errors for each species across sites in each park and across all parks, lists of species that are detected at 95% confidence for each park by the survey method, covariate analyses, and PAO trends with confidence intervals. If an information-theoretic approach is used to compare various models of occupancy and detectability, models will be considered exploratory rather than confirmatory (Burnham and Anderson 2002). These in depth trend analyses will be performed every five years on a per-park basis, and include some regional pooled analyses covering larger geographic areas to increase power (i.e., several parks, and/or non-NPS data, included).

For long-term analyses, a master matrix of species by site, compared through time, will be used to detect any concomitant changes in occupancy patterns across species. Comparison of these matrices through time will allow us to identify apparent turnover in site occupancy (or lack of detection). For example, if a significant proportion of species are consistently not detected at a water body (or collection of water bodies), and these absences are not explained by any weather or related conditions, increased monitoring at the site, or an exploration of other possible explanatory factors, may be merited. Investigations will also seek to determine which variables most strongly affect detectability and presence of each amphibian species. Figures may include trend charts for PAO, maps of sites, annual temperature and precipitation of the current and last 30 years at nearby weather stations, and GIS coverages of any other spatially explicit stressor (e.g., roads). Tables may include parameters of monitoring effort and PAO with confidence intervals and standard errors for each park. Text may also include noteworthy anecdotes or observations.

In long-term analyses, model updating through an information-theoretic approach (Burnham and Anderson 2002) will be performed to see whether occupancy and detectability are changing over time. PAO for each species for each year will be analyzed over the long-term using models designed for this purpose (Mackenzie and Royle 2005) and available in the program PRESENCE. Trends for PAO will be compared with any trends in the numbers of individuals observed per species per site per year, perhaps by incorporating calling indices into PAO (Royle 2004).

Relatively brief annual reports will be provided to each park, including: the total sampling effort (# days, # samples, # sites), descriptions of locations monitored and environmental data collected,

lists/tables of species detected, and outlines of any departures from initial sampling strategy imposed by logistical, equipment, or other constraints.

We have developed a dual approach data analysis protocol (SOP #7), which provides two sets of survey results for modeling purposes. The automated process analyzes hundreds of samples at each site in each year, with recognizers currently available for most species. The manual process analyzes 30 samples at each site in each year, and includes scoring for a Call Index Value (maximum call intensity), which can be analyzed as a measure of abundance. The time required to run a single recording through a Song Scope recognizer file is significantly less than the actual recording time, and files can be batch processed for significant time savings. Similarly, the manual screening process can evaluate 30 samples in <2 hours. Under our sampling design, each ARS will annually record approximately 2,000 5-minute samples (166 hours, ca. 18 GB), and each park with 10 ARS units will produce approximately 20,000 samples per year (1,660 hours, 180 GB). The time required for data management, analysis and reporting will be quantifiable after a trial period. See SOP #7 for additional information on data analysis, and SOP #8 on reporting.

4.5 Data Archival Procedures

GLKN's general strategy for data archiving is described in GLKN's Data Management Plan and appendices (Hart and Gafvert 2006). Guidance specific to this amphibian monitoring protocol is presented here. Data archiving serves two primary functions, it provides a source to retrieve a copy of any data set when the primary dataset is lost or destroyed, and it provides a data record that is an essential part of the QA/QC process (Gafvert and Casper 2018).

Original data are always archived. Original data for any printed forms are either the physical datasheets or exact and complete digital copies of the forms that capture all entries and notations. The unedited files are the original data for digital data.

All digital data will have a duplicate file created at the earliest opportunity and there must always be at least two complete copies of the amphibian monitoring dataset. This includes digital replicas (scanned versions) of any hard copy data sheets or maps. Digital field data that are entered directly into a field computer or collected from a data logger, must be backed up to a second medium each day. Where possible, data files should be duplicated in the field, with at least one copy being stored in a solid state memory product (for example, CF cards and SD cards), which are less susceptible to damage from physical shocks than is a mechanical hard drive. The data files on field computers and loggers must not be erased until verification of the duplicate storage media. Field files should only be deleted when memory space is needed for new data collection and then only when backup files have been verified on another storage medium (second hard drive).

GLKN's master version of the amphibian monitoring Access database is maintained on a central server in the Ashland Office that is backed up daily, and backed up off-site weekly. Complete details of the GLKN Server archiving procedure are found in the Infrastructure chapter of GLKN's Data Management Plan.

5.0 Personnel Requirements and Training

5.1 Roles and Responsibilities

Executing this protocol will depend, in part, on having sufficient GLKN staff and on participation by parks and other collaborators. The expected collaboration is for GLKN to work with individual parks to implement this protocol at APIS, GRPO, INDU, ISRO, MISS, PIRO and SLBE; while the ARMI TWGCRN protocol is implemented by USGS at SACN and VOYA. In this scenario, GLKN will monitor annually in seven park units, and USGS-ARMI in two. Data will be shared for independent analyses by NPS and USGS. Contractors may also be used for some of the roles listed below.

5.1.1. GLKN program manager

The role of the network's program manager is to serve as a liaison among the protocol collaborators, other monitoring partners (e.g., states and provinces), park staff, network staff (field personnel, data manager), and other GLKN monitoring program managers. The program manager is responsible for administering and coordinating the overall program, keeping the network coordinator apprised of progress, and coordinating logistics and information transfer with park staff. Specific responsibilities of the program manager include the following:

- Ensure timely field data collection.
- Ensure availability of equipment and supplies.
- Develop a training program for field personnel.
- Develop and document initial and changes to the protocol and standard procedures for collection and management of field data.
- Coordinate information transfer with park staff.
- Develop quality assurance and quality control measures for the project. Supervise staff training and conduct quality assurance checks of field sampling techniques.
- Supervise or perform data entry, verification, and validation.
- Conduct or supervise routine data analysis and reporting.
- Serve as the main point of contact concerning data content.

The program manager will also work closely with the data manager to:

- Develop data verification and validation measures for quality assurance.
- Ensure staff is trained in the use of database software and QA/QC procedures (Gafvert and Casper 2018).
- Coordinate changes to data forms and the user interface for the project database.
- Identify whether sensitive information should be omitted prior to distribution.
- Ensure regular archiving of project documentation, original field data, databases, reports and summaries, and other products from the project.

- Define how project data will be transformed from raw data into meaningful information and to create automated data summary procedures.

5.1.2. Park personnel

The role of park personnel is to conduct all field work related to the monitoring project. Training and safety are addressed in SOP #2. Responsibilities for field personnel include:

- Completing/passing any needed training for deployment and use of ARS equipment and any ancillary sensors.
- Preparing and maintaining equipment during the season and cleaning, storing, and inventorying equipment after field work has been completed.
- Deploying equipment.
- Checking equipment (e.g., ARS units) during the field season to ensure they are working properly.
- Collecting equipment and survey data (field recordings) at the end of the field season.
- Maintaining accurate field and office notes.
- Ensuring that all quality assurance and quality control procedures are implemented.
- Regularly communicating progress and accomplishments with the program manager at each park unit, and reporting any deviations from sampling protocols.
- Uploading, entering, and verifying data into databases as required.
- Documenting important details of all data collection, including explanations of all deviations from standard procedures.

5.1.3. Data manager

Data management for this monitoring effort will be the shared responsibility of the data collectors first, then the program manager, and finally the network data manager. Field personnel, with oversight from the program manager, are responsible for data collection, data entry, data verification, and validation. The data manager is responsible for data archiving, data security, dissemination, and database design. The data manager, in collaboration with the program manager, also develops data entry forms and other database features (as part of quality assurance) and automates report generation.

5.1.4. Data analysis and reporting

These tasks will most likely be contracted by the network to researchers familiar with the processes.

5.2 Training Procedures

Training needed for deploying, maintaining, and collecting ARS and sensor equipment is minimal. Prior to deploying equipment, field personnel must complete requisite vehicle and first-aid safety training. Personnel must understand the use, calibration, and maintenance of all equipment described in this protocol. Training may consist of self-testing of equipment according to provided manuals and procedures, with practice data sets for uploading and downloading. Personnel who were trained in previous seasons will participate in a review of all methods and techniques.

Training procedures will include:

- Relations, roles, and responsibilities with respect to park staff.
- Basic concepts and field sampling techniques.
- Review of all SOPs for project.
- Calibration, operation, and maintenance of all field equipment.
- Specific techniques for data collection and backups to laptop and desktop computers.
- Safety

6.0 Operational Requirements

6.1 Annual Workload and Field Schedule

The annual workload and schedule for amphibian monitoring is contingent upon phenology, weather conditions, and other GLKN monitoring activities. Park staff and/or a GLKN contractor will deploy ARS and sensor units at ten permanent sampling sites each spring shortly after ice-out, or before ice-out if delayed starts are used. Park staff and/or a GLKN contractor will perform at least one follow-up check to ensure proper operation of equipment, and collect equipment within 2 weeks of recording sessions concluding (14 weeks after deployment). Depending on funding levels, park staff and/or a GLKN contractor may also perform spot checks during the sampling season, along with supplemental human listening sessions to compare to automated recordings. Generally speaking, deployment will be in March and April, and final equipment collection in July-August, depending on the park latitude and local weather phenology. Scheduling of field work will depend primarily upon weather and the phenology of calling amphibian species. The GLKN deployed initial ARS units in INDU, SLBE and PIRO in 2013, in MISS and APIS in 2014, and in GRPO and ISRO in 2015. In future, the program may expand by adding additional ARS units if funding and workloads permit. Training files for developing and testing automated recognizers were also obtained at other sites in the western Great Lakes by the investigators from 2010 through 2016.

6.2 Equipment Needs

Seventy-five Song Meters (model SM2+) with external temperature sensors were purchased by GLKN in 2012. Subsequent purchases for new or replacement equipment will consider newer models, such as the SM3 or SM4 (SM2 is discontinued). Each ARS will annually record approximately 2,000 5-minute samples (166 hours, ca. 18 GB) per year. Most frog vocalizations are under 4 KHz and need only be sampled at 8 KHz. At a sampling rate of 8 KHz in stereo, sampling should consume 32,000 bytes per second, so each 5-minute sample recording should require approximately 9,375 KB. This translates to ca. 18 GB of data per ARS per year. Total storage capacity for transfer and archive media will therefore require 180 GB per park per year. Each park will produce approximately 1,660 hrs of recordings annually, or 10,790 hrs and 1.17 TB for all seven parks combined each year if the program is fully deployed.

6.3 Startup Equipment Costs

Initial acquisition of 75 Song Meters with external temperature sensors (SM2+ Terrestrial Acoustic Packages and microphones) cost $55,809 in 2012. High capacity flash cards are available up to 128 GB. For this sampling design, each Song Meter will require 18 GB capacity, but we recommend using two 32 GB cards in each Song Meter, which allows for continuous recording, should one fail. Appropriate flash cards should cost about $10 each (× 75 × 2 = $1,500). Each Song Meter requires four standard alkaline D-size batteries at approximately $15 for 24 ($0.63 each or $2.52/Song Meter × 75 = $189.00/year). Each Song Meter also requires two standard alkaline AA-size batteries at approximately $20 for 40 ($0.50 each or $1.00/Song Meter × 75 = $75.00/year; this cost is absent for SM3s). All batteries should be replaced annually. Additional costs may include mounting screws, an external temperature logger (ca. $50 each), posts if the units cannot be attached to trees, and other field supplies normally already in hand with park personnel (e.g., rules, hip waders, thermometer;

many of these items may already be available in parks for other uses). Dedicated storage, transfer, and archival media costs may also need to be determined. The current network investment in equipment to get the program up and running was approximately $60,000. Ongoing annual expenses will be primarily for batteries, and replacement of damaged microphones or Song Meters.

6.4 Personnel and Travel Costs

No additional staff are anticipated to be hired by GLKN or the parks to implement this protocol. Travel costs are expected to be minimal, as park staff will deploy and retrieve the equipment.

6.5 Contracting Costs

GLKN will request bids for contracting services to help implement portions of this protocol, primarily for data analysis and reporting. The cost of the contract may vary across years, depending on factors such as the degree of data analysis required and the type of resulting report. The cost of the contract is estimated at $15,000 per year at this time.

6.6 Procedures for Revising and Archiving Previous Versions of the Protocol

As the amphibian monitoring program matures, revisions to both the protocol narrative and specific standard operating procedures (SOPs) are likely. Documenting changes and archiving copies of previous versions are essential for maintaining consistency in the collection of data and for appropriate interpretation of data summaries and analyses.

This protocol narrative includes a general overview of the history and justification for doing the work and an outline of the sampling methods. The SOPs are more detailed step-by-step procedures of methods. Whenever an SOP is revised, the corresponding section of the protocol may or may not need to be revised depending on the type of change. All old versions of the protocol narrative and SOPs will be archived.

The steps for changing either the protocol narrative or the SOPs are outlined in Procedures for Revising the Protocol – SOP #9. Each SOP contains a Revision History Log that should be filled out each time a SOP is revised to explain why the change was made, and to assign a new version number to the revised SOP. The new version of the SOP or protocol narrative should then be archived in the appropriate folder of the GLKN database structure.

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